The Journal of Parasitology

A Quarterly Devoted to Medical Zoology

Volume III

The Journal of Parasitology

Volume 3 SEPTEMBER, 1916 Number 1

CONTRIBUTIONS TO THE STUDY OF PARASITIC PROTOZOA. III.

NOTES ON MYXOSPORIDIA FOUND IN SOME FRESH-WATER FISHES OF JAPAN, WITH THE DESCRIPTION OF THREE NEW SPECIES (WITH FOUR TEXT FIGURES)

ROKUSABURO KUDO

THE IMPERIAL SERICULTURAL EXPERIMENT STATION, NAKANO, TOKYO

In a former paper (1915) I described from a morphological as well as developmental point of view, a new species (*Myxobolus toyamai* Kudo) from the branchial lamellae of a carp. The present paper is the result of study upon Myxosporidia found since that time. I am now working on the life-histories of the new species described below with the hope of reporting them later.

1. *Myxosoma dujardini* Thel.

Eight cysts (the largest being 200μ in diameter) of round shape, were found in the branchial lamellae of a carp 23 cm. in length. The seat of the parasite was, as in *Myxobolus toyamai,* the connective tissue of the gill-filament.

The results of observations upon the spore coincide for the most part with the description of Thélohan (1895). I wish, however, to give here details about the polar capsule and the polar filament, as Thélohan failed to mention them: length and breadth of the polar capsule 6-7μ and about 2μ, respectively, and the length of the polar filament about 70μ.

2. *Zschokkella acheilognathi* n. sp.

Vegetative form. Large ones are generally visible to the naked eye as small, opaque, more or less regular, usually subspherical masses, occupying various parts of the gallbladder and especially of the gall-duct (Fig. 1). The size varies with age up to a maximum length of 720μ by a breadth of 550μ, and the thickness of one individual is about uniform throughout, but in many specimens it differs from 5 to 30μ according to the size of the myxosporidium. Their bodies are very flexible and easily doubled up, representing, in sections, various forms. The vegetative stage in sections resembles much that of *Sphaeromyxa*

hellandi, observed by Auerbach (1912), both in form and structure. The body is colorless in both young and old. In its fresh condition the protoplasm can be seen to be clearly differentiated into finely granulated reticular ectoplasm and greatly vacuolated endoplasm. In the younger form (15 to 30μ in greatest diameter) lobose pseudopodia are well developed. The myxosporidium moves about more or less actively in the bile by the constant emission of pseudopodia. No clear evidence of the active emission of pseudopodia exists in older individuals. The ectoplasm of some more advanced specimens shows in section two structures; the outer layer, comparatively thin but uniformly about 2μ in thickness, presents very fine striations, while the remaining part is finely alveolated, having an average thickness of 6 to 8μ.

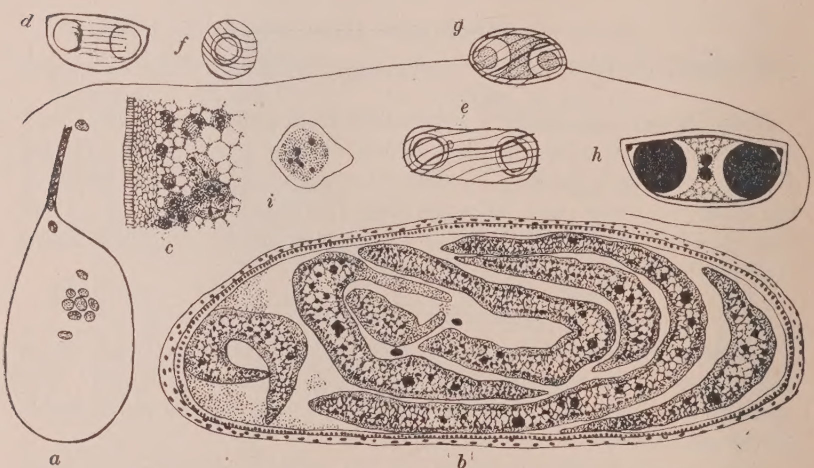


Fig. 1.—*Zschokkella acheilognathi* n. sp. *a,* Gall-bladder of Acheilognathus with many myxosporidia, $\times$ 10; *b,* oblique cross section of the infected gall-duct, $\times$ 200; *c,* part of the cross section of the parasite, showing the differentiation of the protoplasm, $\times$ 1,000; *d,* to *g,* spores: in *g,* filaments are extruded, $\times$ 1,500; *h,* stained spore, about $\times$ 2,250; *k,* young myxosporidium, $\times$ 1,000.

Auerbach (1910b) seems to have observed similar structure in the outer layer of the ectoplasm in *Sphaeromyxa hellandi,* sketching a surface view of an individual fixed in formol. It takes stains much more deeply than the endoplasm, so that in sections it shows clear differentiation of the protoplasm much better than was shown in *Myxobolus toyamai.* The endoplasm contains vegetative nuclei as well as generative nuclei in several stages of spore formation. It is polysporous, according to the observation made up to the present time. In this regard, it is quite different from *Zschokkella hildae,* in which, after Auerbach (1910a), single and double spore formation occurs.

Spore. Generally oval with round poles, very often more or less

hemispherical, somewhat attenuated symmetrically at both ends of the flat side of the spore. Several modifications in form and size, however, are also found in the present case as in other forms. It is usually 10 to 14μ long by 6 to 7μ wide. The thickness is almost equal to the width. Shell bivalve; the line of junction being oblique to the longitudinal axis of the spore. Parallel to the line of junction, fine striations run longitudinally on the spore coat. A polar capsule, round in shape, with a diameter of 2 to 3μ, at each end of the spore, takes stains very deeply. Polar filaments were easily extruded by the application either of mechanical pressure or KOH-solution, and are well stained after my method (1913). The fully extruded polar filaments were 65 to 70μ long.

Habitat. This species is found quite abundantly in the gallbladder of *Acheilognathus lanceolatum* Temm. et Schl., commonly found in brooks in the vicinity of Tokio. Out of twenty-four fish (8 to 12 cm. long) examined in May, 1915, twenty-one were found to have harbored the parasite; thus the rate of the infection rises above 80 per cent. Matured, large vegetative forms were very often found in great numbers in the gallduct, while in the gallbladder of the same host I could find only a small number of isolated spores. Klokacewa (1914) described a somewhat similar form of spore from *Carassius vulgaris,* without finding the vegetative form. The species in question, though its vegetative form differs apparently from that of *Zschokkella hildae*, seems to belong to that genus. Up to the present time two species of the genus have been reported, that is, *Zsch. hildae* and *Zsch. nova,* since Auerbach (1910) created it. The former apparently differs from the present form in several points. Now the dimensions of the spore of the latter seem to correspond very nearly to the myxosporidium in question. As it lacks, however, all other details, it is impossible to make accurate identification, so I treat this species as a new one, calling it *Zschokkella acheilognathi.*

3. *Myxobolus fuhrmanni* Auer

Isolated spores of this form occur very often in the bile of the loach (*Misgurnus anguillicaudatus*). The vegetative form has not yet been found by me. The description of the spore by Auerbach (1909) coincides well with my observations, except that I found the thickness of the spore coat to be uniform, whereas Auerbach's observation was in effect that the shell is especially thick at the posterior end of the spore. I wish to mention that the length of the polar filament in the present case is 100μ. Nearly 50 per cent. of the said fish, examined in September, 1915, were infected by this Myxobolus. In all cases, however, the infection seems to be carried to a very slight degree.

4. *Myxidium* sp.

This form, together with the following two new species, are also found in the gallbladder of the loach. The vegetative form has not yet been observed. Two per cent. of the fish studied in September, 1915, were infected. The spores are mostly found separated from each other, floating in the bile. One side of the spore is, in most cases, more convex than the other. The sporoplasm usually occupies the whole inner space of the spore, except the polar capsules, and shows fine granulations in the natural condition as in the case of *Myxidium giardi* or *Myxobolus pfeifferi,* and also shows a fine alveolar structure in stained preparations. It contains two nuclei of almost equal size. Shell bivalve, the line of junction of which is straight. On the surface of the shell, fine striations run longitudinally parallel to the line of junction. The dimensions are: length, 15 to 18μ, breadth 6 to 7μ, length of polar capsule 7 to 8μ, and that of polar filament 60 to 70μ.

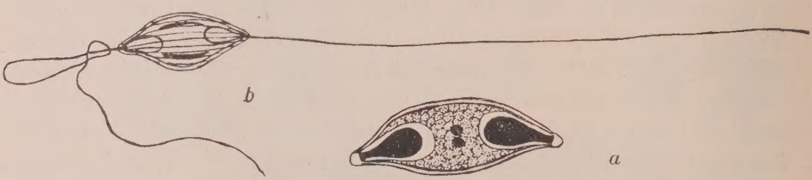


Fig. 2.—*Myxidium* sp. *a,* stained spore, $\times$ 1,750; *b,* spore with extruded polar filaments, $\times$ 1,000.

Ishii (1915) recently described a new species, *Myxidium anguillae,* from an eel. The form in question apparently differs from any of the species reported up to the present time. I hope to identify the species after studying it more closely.

5. *Chloromyxum misgurni* n. sp.

Vegetative form. Mostly round, often of irregular form. From a side view it assumes a semicircular shape. From the more or less flat surface many fine root-like pseudopodia extend. They are more clearly visible in younger specimens, where the spore formation has not yet begun. There is no clear differentiation of protoplasm. It is finely vacuolated on the whole. With the pseudopodia, the myxosporidium probably creeps along the surface of the epithelial layer of the gallbladder, so that many individuals in different stages of development are found in section preparations, closely attached to the epithelial cells with their peculiar pseudopodia. The size varies with age, the largest being 50μ in greatest diameter, with the maximum thickness of 20μ. Individuals with six to eight spores are of common occurrence; those with twelve to sixteen spores, however, occur rarely, and it is very seldom that only two spores are found in one myxosporidium.

Spore. Spherial, slightly attenuated at the anterior end. Shell

bivalve; the line of junction straight. Parallel to the ridge which marks the line of junction very clearly, run fine longitudinal striations. Four polar capsules are situated in the anterior end. In the finely granulated sporoplasm two nuclei of equal size are found. The dimensions of the spore are: length, 8 to 9μ, breadth 6 to 7μ, thickness 5 to 6μ, length of the polar capsule 2 to 3μ and that of polar filament 28 to 35μ.

Habitat. In the gallbladder of *Misgurnus anguillicaudatus* Cantor.

Of the fish examined in September, 1915, 73 per cent. were found to be infected.

Of forms known up to the present time, *Ch. fluviatile* (Thélohan, 1895) seems to be nearest in size and form of the spore and seat of infection to the Chloromyxum mentioned. The form and size of the

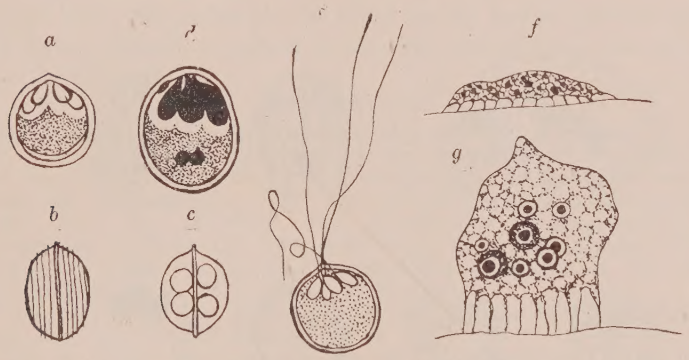


Fig. 3.—*Chloromyxum misgurni* n. sp. *a,* front view; *b-c,* side view of the spore in natural condition, × 1,750; *d,* stained spore, × 2,625; *e,* spore with the extruded polar filaments, × 1,750; *f, g,* two young myxosporidia in section, × 1,750.

pseudopodia and the structure of the spore of the type studied differ from that described by Thélohan. Therefore, I propose to name this *Chloromyxum misgurni.*

6. *Chloromyxum fujitai* n. sp.

Vegetative form. Mostly round, sometimes irregular. There is no clear differentiation of the protoplasm. The endoplasm is highly vacuolated, the ectoplasm being hardly visible. The largest one was 40μin diameter. They float about in the bile, so that in sections of infected gallbladder they are found in the gall apart from the epithelial layer, by which fact we can easily distinguish them from *Chloromyxum anguillicaudati,* even when both forms occur in the same gallbladder. Disporous and polysporous, with up to eight spores.

Spore. Circular in general; often attenuated at the anterior end Shell bivalve; the line of junction not being straight, but very thick.

The shell has peculiar thick ridges running longitudinally on the surface. Near the anterior end of the spore two small circular markings are clearly visible, in preparations well stained with Heidenhain's iron hematoxylin, one on either side of the line of junction, from which the markings recede on both valves. These two circular markings are not the exits for polar filaments, because it is clearly shown in preparations stained with Giemsa's solution that the four polar capsules have their independent exits. The form of the spore takes different aspects by the presence of the characteristic markings resembiing partially those of *Hoferia cyprini* (Doflein, 1898) and *Chloromyxum koi* (Fujita, 1913). In optical cross-section, the spore represents an outline quite like a cog-wheel with twenty to twenty-two ridges, including the widest ridges, which mark the line of junction of the shell. The

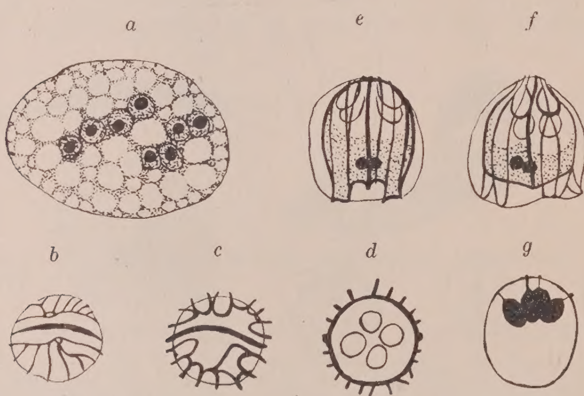


Fig. 4.—*Chloromyxum fujitai* n. sp. *a*, young vegetative form, $\times$ 1,750; *b*, *c*, anterior (*b*) and posterior (*c*) view of a stained spore. $\times$ 1,750; *d*, optical cross section of the same spore as the above, $\times$ 1,750; *e*, *f*, side views of stained spores, $\times$ 1,750; *g*, side view of stained (Giemsa) spore, $\times$ 1,750.

thickness of the ridges varies regularly. The thickest ones are located where a plane perpendicular to that of junction cuts the shell longitudinally, others decreasing in thickness as they approach the line of junction. Four polar capsules occupy the anterior half of the spore. The sporoplasm contains two nuclei of almost equal size. The dimensions are: length 10 to 12μ, breadth 8 to 10μ, length of polar capsule 2 to 3μ, and that of the polar filament 23 to 30μ.

Habitat. In the contents of the gallbladder of *Misgurnus anguillicaudatus*. The occurrence is much rarer than that of the former one, showing about 5 per cent. in September, 1915.

Of all descriptions from this genus that of Fujita (1913) alone described similar markings of the spore of *Chloromyxum koi* from the gallbladder of the carp. However, we find a great difference

in form and in the number of ridges. There is also a great difference between the size and structure of the spore, and in the number of the spores found in a vegetative form. So I think this species is a new one. In honor of Dr. T. Fujita, who was the first to study myxosporidia in Japan and to discover the spore of this type, I give the name *Chloromyxum fujitai*.

Multiple infection of the above-mentioned four species takes place very often.

Concerning the pathological effects, I have but little to report. No visible external change could be noticed in any of the infected fishes. But, as in the case of Acheilognathus, we found often that the gall-duct had been filled up with a great number of the Zschokkella (Fig. 1), therefore it is certain that the secretion of the bile into the duodenum must be greatly disturbed. Such is the case with the highly infected loach, the gallbladder of which has an opaque appearance. It is, however, difficult at present to state the real effects of the parasite upon the host.

References Cited

Auerbach, M. 1909. Bemerkungen über Myxosporidien. Zool. Auz., 34: 65-82. 1910. Biologische und Morphologische Bemerkungen über Myxosporidien. Zool. Anz., 35: 57-63. 1910a. Die Sporenbildung von Zschokkella und das System der Myxosporidien. Zool. Anz., 35: 240-56. 1910b. Cnidosporidienstudien. Zool. Anz., 35: 767-77. 1912. Studien über die Myxosporidien der Norwegischen Seefische und ihre Verbreitung. Zool. Jahrb., Anat., 34: 1-50.

Doflein, F. 1898. Studien zur Naturgeschichte der Protozoen. III. Ueber Myxosporidien. Zool. Jahrb., Anat., 11: 281-350.

Fujita, T. 1913. On a new species of Chloromyxum from the gall-bladder of the carp. Annot. Zool. Japan., 8: 257-59.

Ishii, S. 1915. Myxosporidiosis of the eel. (Japanese.) Trans. Tokyo Zool. Soc., 27: 372-82.

Klokacewa, S. 1914. Ueber die Myxosporidien der Karausche. Zool. Anz., 44: 182-86.

Kudo, R. 1913. Eine neue Methode die Sporen von *Nosema bombycis* Nägeli mit ihren ausgeschnellten Polfaden dauerhaft zu präparieren und deren Länge genauer zu bestimmen. ZXool. Anz., 41: 368-71. 1915. Contributions to the Study of Parasitic Protozoa. II. *Myxobolus toyamai* n. sp., a new myxosporidian Parasite in *Cyprinus carpio* L. (Japanese.) Trans. Tokyo Zool. Soc., 27: 517-23.

Thélohan, P. 1895. Recherches sur les Myxosporidies. Bull. scient. France et Belg., 26: 100-394.

NOTES ON TWO FREE-LIVING LARVAL TREMATODES FROM NORTH AMERICA *

Henry B. Ward

The life history of parasitic worms has always been a subject of especial interest and no part of it commands more careful attention than that which deals with the brief stages of free existence, for here is the point at which the organism effects its transfer from one host to another. It is only in the few highly specialized types that the parasitic habit endures in unbroken succession from host to host and the parasite is transferred by the agency of the organism in which it is living. This is the case with the *Plasmodium malariae* and all other parasites transferred by a blood sucking host to a new environment in which usually if not always a new generation is developed. It is also the case with the trichina and other encysted parasites which are acquired by a new host thru its carnivorous habit and which develop into a new form or stage of the life history in this new host rather than as a new generation.

In the simpler cases, however, the parasite abandons at intervals the parasitic mode of life and adopts a free-living habit for a stage of its existence or for a generation that alternates with the parasitic type. The free-living generation or stage is of especial interest because it affords the opportunity for the infection of a new host and also furnishes a point of attack in the life cycle which is vulnerable so that the readjustment of environmental conditions may block the transfer and prevent the infection. Such a readjustment may result from the natural operation of external forces, as when a very dry season eliminates the small ponds or swamps in which the free stage develops and thru which it secures means of transfer to the new host. Or the change may be brought about by the introduction of hygienic regulations that are drafted to prevent the parasite from reaching its new host, as the installation of a new sewage system may divert the human feces with tapeworm eggs from the lake into which they were formerly discharged and thus prevent infection of the fish host in that lake with the bladder stage of the fish tapeworm (*Dibothriocephalus latus*); consequently the spread of the tapeworm in the human host is checked.

---

* Contributions from the Zoological Laboratory of the University of Illinois, No. 71.

Attention has been forcibly directed in the past year to the free-living stages of trematodes by the work of Japanese and European investigators on the life history of the Lungfluke (Paragonimus) and the Bloodfluke (Schistosoma). As a result of these studies it is possible now for the first time to draw a reasonable picture in outline of the development of these forms. Almost nothing has been ascertained concerning the free-living stages of such parasites in North America. A few observations were made years ago by Leidy and recently Cort has published a careful study of some larval trematodes; but together these cover only a few of the many North American species of fluke. The importance of placing on record all data leads me to print here observations made some years ago although they are yet unfortunately incomplete. For the satisfactory interpretation of these observations a preliminary statement may be made here regarding the process of development as found in the trematode.

Two free-living stages recur in the development of most flukes. From the egg develops a small ciliated larva, designated a miracidium by Braun, which is evidently dependent upon water for its distribution. It remains within the shell until it has reached its full development; thereafter contact with the water is sufficient to open the shell and bring about the escape of the embryo. Active migration through the water permits it to reach and infect the secondary host, a mollusk. It is a somewhat striking fact that in spite of the constant and abundant production of eggs and embryos, no records that I have found note the occurrence of such embryos in plankton or other fresh-water collections. The absence of records can not be attributed to the small size of the miracidia since other even more minute objects fall constantly within the ken of the microscopist engaged in the study of fresh-water organisms, and some that are apparently very difficult to detect have been studied and described in detail. It may be due to the extreme delicacy of the larvae which are thereby readily subject to accidental destruction. Certainly they go to pieces almost as soon as they are collected.

The second free-living stage in the life history of the trematode comes when the cycle of development within the mollusk is completed and the transfer to the adult host takes place. This transfer occurs in the cercaria stage and of course may be direct if the mollusk is eaten by a suitable host. Yet one may safely infer that this is not the usual method since most cercariae are so well adapted to a free aquatic existence. The ordinary cercaria possesses a well developed swimming organ in the tail which characterizes this stage and is cast off when the larva reaches a new host or a place of encystment, as the case may be.

This swimming tail is reduced in a few types and absent only very infrequently.

In other cases the tail is not only present but powerful and displays various modifications, such as bristles, folds, branches, lateral membranes, etc., that increase its functional value. No one who watches living cercariae in the laboratory under experimental conditions can doubt that they are robust swimmers and naturally depend on that method for their transfer from the mollusk to the final host. When infected snails are kept in an aquarium jar on the laboratory table, the cercariae swarm out voluntarily at certain times in great numbers and in many cases can be seen with the unaided eye swimming actively about in the water. They do not confine themselves to the sides or bottom of the vessel but seem to seek equally the open water and in general to conduct themselves under such circumstances like other plankton organisms: protozoa, rotifers, and entomostraca in the same aquarium.

These cercariae are large objects among the microscopic aquatic organisms; they are produced in great abundance and infected mollusks are also abundant and widely distributed. And yet there are almost no records of the occurrence of cercariae in the voluminous reports on fresh water plankton and aquatic life. I am at a loss myself to explain this condition. I have seen them many times in fresh water collections, but only when the material was examined very soon after it was taken in the net. Usually the number of specimens secured was too limited to permit of satisfactorily determining the structure and relationships of the form. In two cases, however, the cercaria was so peculiar as to justify this record of its occurrence even though the description is incomplete in certain respects.

CERCARIA ANCHOROIDES *nov. spec.*

The first species to which attention is called was abundant at Lake St. Clair in 1893 and was described very briefly in a preliminary report (Ward, 1894), as follows:

In the tow was found but one helminth, a form which challenged attention the first of our stay. It is a free-swimming Cercaria, closely allied to *C. mirabilis* Braun, having a prominent tail terminating in two flat blades at right angles to the main body. The distome is enclosed within the tail of the Cercaria which has then more or less the appearance of an anchor with wide flukes. From one to four of these were taken from both top and bottom tow every day from July 27 to August 5 when it suddenly ceased to be found. Efforts to find the intermediate and primary host were alike unsuccessful. This form differs consid-

erably in size from those described by R. R. Wright and M. Braun and is probably a new species.

The cercaria attracted attention by its very active movements, which were so peculiar that it could be readily picked out from other material in a glass dish. While found in collections from the surface as well as in those taken by a runner net and representing thus the fauna near the bottom, yet it was three times as frequent in the latter as in the former. This probably indicates that the cercariae were discharged from snails or other mollusks living on the bottom in the region where the collections were made.

Viewed under a lens the organism appeared as a minute object shaped like a hammer or anchor and swam through the water by a succession of violent jerks. The body moved with the flukes of the anchor in advance and propelled itself by throwing the flukes alternately right and left. In this movement the flexure occurred in the stem of the anchor about one third of the distance from the flukes to the head, and the blades did not move separately but maintained constantly the same relation to each other and to the adjacent part of the stem. The motion recalled distinctly the sweep of a double headed paddle as it is passed from the one side of a canoe to the other.

A more careful examination showed that the stem of the anchor was flat and also the flukes which extended from it nearly at right angles but were curved a little near the outer end. The head of the anchor appeared, however, nearly round, being enlarged and enclosing a small object which lay in a clear, fluid-filled chamber at the extreme head end of the stem (Fig. 2).

The stem of the anchor measured about 2 mm. in length and the flukes were each 0.53 to 0.6 mm. long, though on account of the curve their tips were only 0.84 mm. apart. The flukes varied in width from 0.24 to 0.34 mm. and the stem was 0.28 mm. in maximum width but was reduced to 0.2 mm. at the region of transition from the flat base to the rounded head. At its widest part near the outer end this region had increased again to about 0.3 mm. in diameter.

Under the microscope one could readily distinguish that the object in the enlarged end was a small distome. It lay with the oral sucker near the apex of the chamber and with the opposite end turned towards the flukes of the anchor. In no case was it coiled, twisted, or crumpled together in life but lay flat and straight with abundant space in the chamber for it to be extended to full length. When the distome lay flat on the slide, the base of the stem and the flukes of the anchor stood on edge, thus the breadth of the distome was at right angles to the flat surface of the stem and flukes.

The living distome was faint sulphur yellow with a reddish brown intestine; the anchor stem and flukes were dark by transmitted light and white or faint yellow by reflected light.

Under the pressure of the compressor the young distome (Fig. 1) was forced out of the sac in which it was contained. It emerged at the extreme tip where there seemed to be a preformed opening and left wrinkled and collapsed a thin walled sac in which it had been enclosed. The base of the stem and the flukes of the anchor remained entirely unchanged and often moved about actively in the water for some time after the distome had escaped, beating alternately right and left in the same manner as before and moving freely through the open water just like those specimens in which the distome was still enclosed in the sac.

The young distome, freed from the sac in the tail, as the anchor should properly be called, measured in life 0.64 mm. in length by 0.288 mm. in breadth. The oral sucker was sub-terminal or ventral, being separated 0.016 mm. from the extreme anterior tip; its diameter was 0.16 mm. and its orifice measured 0.048 $\times$ 0.064 mm. The ventral sucker though conspicuous was a little smaller than the oral, measuring 0.128 by 0.144 mm. and being separated from the anterior tip by a distance of 0.27 mm.; its orifice measured 0.04 by 0.072 mm. The pharynx was 0.064 mm. in diameter, and in some cases even a little longer.

The intestine was large, broad, wavy in outline, and filled with a dark reddish brown fluid containing numerous highly refractive granules. The main duct of the excretory system extended from the posterior tip to the acetabulum and two longitudinal trunks were conspicuous on the right and left sides of the worm, outside the intestinal crura. They were not straight but much twisted or thrown into short heavy wavy loops from which fine branches extended towards the margin and gave rise to still finer branches. In one specimen a transverse connection was demonstrated just anterior to the acetabulum extending from the median posterior trunk to the left longitudinal trunk. It was also noticed that the right canal in the same worm was certainly larger near the center of the body than near either end. This condition would indicate that a connection existed at the center in this tube also. One would not go far astray in interpreting the median posterior stem as the bladder from the apex of which near the acetabulum branches extended right and left to divide again near the margin on each side of the body into anterior and posterior trunks.

On the ventral surface of the body appeared a transverse slit just 15μ in front of the anterior margin of the acetabulum in the living specimen, or halfway between it and the oral sucker in a contracted alcoholic specimen. While the ducts connected with it were not yet

developed or at least demonstrable there seems little doubt that this represents the genital pore. In the center of the body behind the acetabulum and between the intestinal crura three faint masses were discernible. The two smaller bodies near each other slightly oblique to the axis and furthest posteriad, are probably the testes and a single larger mass between the testes and the acetabulum is no doubt the ovary. These structures were faint, especially the ovary, and the connecting ducts of the genital system were not yet apparent. The three bodies differ slightly in size and location in different specimens.

The sac at the head of the anchor stem had on the outer surface peculiar vacuolated wart-like protuberances. These may have been sensory structures. In the outer wall were also two sets of fibers—probably muscular—of which those in the transverse series are closer together and very regular whereas the longitudinal fibers are neither so abundant nor so regular. The wall of the sac adjacent to the cavity consists of a thin epithelial layer.

In 1885 R. Ramsey Wright found a single specimen of a similar form swimming actively in a fresh water aquarium at Toronto. He published a brief note (Wright, 1885) in which the form was interpreted as a free-swimming sporocyst. Leuckart to whom the specimen was sent with notes and a sketch published (1886) a more extended description with a copy of Wright's drawing. From this account it is easy to determine that the form is very similar to that described above but not identical with it. Both the length which is "nearly 1 mm." (Leuckart) as against 2 mm. in the new species and the form of the tail as well as of the young distome indicate the specific difference of the two types. As appeared later both Wright and Leuckart were in error in the interpretation of this organism which is not in any sense a sporocyst but a true cercaria although with an unusual type of tail. No name has ever been given to this species, which may be designated *Cercaria wrightii* nov. spec. According to the sketch published by Leuckart (1886:102) the distome fills three fifths of the stem of the anchor, the flukes are nearly straight and together two thirds as long as the stem, and the genital organs of the distome form a solid rod-like mass located above the acetabulum but partly preacetabular and partly postacetabular. The approximate measurements of this form taken from the drawing are, total length 0.75, maximum width 0.133, length of flukes 0.533, breadth of flukes 0.1, length of distome 0.45, breadth of distome 0.1, diameter of oral sucker 0.041, of ventral sucker 0.075 mm. Leuckart speaks of the movement of this species as produced by flapping the two wing-like flat fins, evidently depending on the notes of Wright, though the latter in his printed notes did not refer to the way which the animal moved.

Later M. Braun (1891) published an extended account of a similar form which he also found swimming in an aquarium. The material came from Kurland. He was able to show that the form was not a sporocyst but a true cercaria in which the tail was developed to form a receptacle at the anterior end for the body of the young distome. He traced the larva back to *Limnaea palustris,* var. *corvus* and found in the lungs of these snails the numerous sporocysts from which the cercariae had come and in which could still be seen all stages in their development. He named this form *Cercaria mirabilis,* and decided that it was certainly different from Wright's species. From his description which agrees in general with Wright's type and with my own, some items may be cited to indicate the differences between the three.

Cercaria mirabilis Braun is 6 mm. long with leaflike, movable wings, 1.5 mm. long. In resting it lies on the bottom with folded wings. In swimming the wings are moved actively from side to side, making the motion resemble that of mosquito larvae. The young distome was an opaque yellow body, lying bent in a space lined by a smooth membrane. Granules of yellow pigment occur abundantly forming a network of lines in the bulb wings, and stalk, and also massed in the intestinal crura. The acetabulum is larger than the oral sucker. The rudiments of two testes and the ovary lie behind the acetabulum.

Since the early stages in the development of *Cercaria mirabilis,* taken from sporocysts, are split-tailed cercariae, Braun regarded this form as the higher differentiation of that type. He compared it with a number of known species but found nothing that furnished a real parallel. He was also unable to suggest to what adult distome this larval form should be assigned.

One further point deserves additional emphasis, namely that the method of swimming adopted by all three of these forms differs widely from that of a typical cercaria. While there are minor differences in movement as already noted, yet all accounts agree in stating that the forms travel with the wings in advance, trailing behind them the stem at the end of which lies the distome in the chamber. The distome is so oriented in these cercariae that the anterior end hangs down or back and the posterior end is pointed in the direction in which the larva is moving. It will be clear on comparing this with the usual cercaria that the orientation is precisely reversed; instead of being pushed ahead by the tail the distome in this case is pulled along after the tail. This is an extremely interesting case of the reversal of functional activity.

CERCARIA GORGONOCEPHALA *nov. spec.*

The other cercaria came from Lake Erie near Put-in-Bay, Ohio. It was taken in a tow from a depth of 4 fathoms on July 23, 1901. Unfortunately only a single specimen was obtained. Under a dissecting microscope the object resembled a writhing mass of serpents tied together by the tails. The diagrammatic representation (Fig. 3) gives, unfortunately, little idea of the actual appearance this object presented in life. The bunch contained not less than 50 separate stalks fastened firmly together at the base, all in active motion with a spiral twist passing in wave-like progression from the base to the outer end of the stalk (Fig. 5). The base of each stalk carried a bulbous expansion which in some stages of contraction appeared to be sharply cut off from the rest of the stalk but at other times graded into the stalk without any distinct boundary. This basal enlargement was thicker walled than the stalk elsewhere and possessed yellow pigment granules that were not found in other parts. The stalk was very mobile, delicate in texture and provided with two irregular longitudinal stripes of granular brown pigment which terminated just short of the outer tip. This stalk which is the region of marked contractile activity, tapered slightly (Fig. 4) to the extreme outer tip where it bore the body of a young trematode that appeared to be an amphistome. About half of the stalks had already lost their attachments; most of these still twisted and vibrated nearly as actively as those that carried the young helminths; but a few were pale in color, appeared empty like dead algal filaments, and were motionless. At times all the stalks rested quietly for a brief period and then suddenly began to be violently agitated together. During this movement it was clear that the stalk alone was active whereas the worm was snapped too and fro like the lash of a whip.

As the stalk vibrated with a coiling or twisting motion, the worm at the end was turned at every angle and it seemed as if the large posterior sucker was mounted on a stalk or at least protruded distinctly beyond the general surface of the body. The general form of the amphistome was oval with a concave ventral surface and the anterior end rolled slightly ventrad (Fig. 6) so that the oral sucker was partly concealed. The body of the worm was white, opaque and almost entirely immobile. No internal organs could be detected either in the attached worms or in those that had been shaken off and lay free in the dish. The posterior end of the detached amphistome bore a distinct projection which indicated the point at which the stalk had been attached.

A somewhat similar form was discovered by Claus in the Mediterranean in 1880 and recorded by Leuckart (1886: 87). It is referred to

in various other places as a Rattenkönigcercaria. It was carefully described by Pintner (1891) under the name of *Cercaria clausii* given it by Monticelli in 1888. Odhner has more recently (1911) shown this to be the larval form of *Phyllodistomum acceptum* Looss. Apart from the peculiar habit that both forms are borne on stalks tied in bunches there is no close similarity between *Cercaria clausii* and *C. gorgonocephala*. The details of structure in the stalk are as unlike as the structure of the two worms.

It is not without some reserve that the species described above is assigned to the amphistomes. The form of the body suggests this connection but the amphistome cercariae thus far known are very different in type. Cort (1915) has described two species from this group very fully and in common with earlier investigators he finds that two eye spots are present in these cercariae. No eye spots were observed in *Cercaria gorgonocephala*. It is possible that the large sucker of this cercaria is in reality not posterior but ventral and that the postacetabular region is yet undeveloped. No definite opinion on the relationships of this form can be given until further material is obtained.

In one minor structural feature this species manifests a striking resemblance to another amphistome cercaria described by Cort. It is clear that the stalk is the homolog of the cercaria tail and as can be readily seen from an inspection of the sketches published herewith this stalk is attached to the young trematode not at the extreme posterior tip so as to form a direct posterior extension of the body, but rather to the dorsal surface just anterior to the end of the body so that the longitudinal axis of the worm when extended lies ventral to that of the stalk. This same relation mentioned by Cort in his text is beautifully illustrated in his sketch of *Cercaria diastropha* (Pl. 3, Fig. 24). The condition exists also in other amphistome cercariae.

One other point deserves especial notice here. While the stalk in this species is in one way an organ of attachment rather than a swimming organ, yet it retains its muscular development and structure relatively unchanged, and in movement vibrates in a manner which suggests to the eye the close similarity to the moving tail of an isolated swimming cercaria. One can then not find a basis for calling this in a real sense a change of function in the organ. The young distome in *Cercaria gorgonocephala* (Fig. 6) is broader and flatter than any yet described in the group of Amphistomata. North American adult amphistomes are only very imperfectly known and I am unwilling at present to venture a conjecture as to the form to which this cercaria belongs.

The two cercariae discussed in this paper manifest an interesting biological similarity. Both differ from the ordinary cercaria in the peculiar development of the tail, which has become so highly specialized

that its original character is not easily discerned. In one case it has become a very efficient organ of locomotion, conspicuous both for its size and its power; in the other case it has given up its swimming function though not its power, and serves to attach the larval worm to others in the group. In both cases the end result is the same: The larva swimming about in the open water forms a most conspicuous object and is readily snapped up by fishes as Braun determined experimentally with *Cercaria mirabilis*. The conspicuous character of both larvae was also shown in their prompt discovery in the tow although they were surrounded by large numbers of other plankton organisms. Their activity and conspicuousness must be helpful in enabling them to reach a suitable host for further development, and such a host will naturally be sought among the fishes of the waters in which the larvae occur.

SUMMARY

A description is given of the structure and activity of two new cercariae of peculiar type captured free in Lake Erie and Lake St. Clair. They are designated *Cercaria anchoroides* nov. spec. and *C. gorgonocephala* nov. spec., and are compared with known European species. Altho such an occurrence must be common, these forms are the first to be taken in open fresh waters.

References Cited

Braun, M. 1891. Die sogenannte "freischwimmende Sporocyste." Centr. Bakt. Par., 10: 215-219.

Cort, W. W. 1915. Some North American Larval Trematodes. Ill. Biol. Monogr., 1: 447-532, 8 pl.

Leuckart, R. 1886. Die Parasiten des Menschen. Leipzig, Bd. 1, Abt. 2, Lief. 1.

Monticelli, F. S. 1888. Saggio di una morfologia dei Trematodi. Napoli, 130 pp.

Odhner, Th. 1911. Zum natürlichen System der digenen Trematoden IV. Die Familie Azygiidae n. fam. Zool. Anz., 38: 513-531, 2 figs.

Pintner, Th. 1891. Ueber Cercaria Clausii Monticelli. Arb. Zool. Inst. Wien., 9: 285-294, 1 pl.

Ward, H. B. 1894. A Preliminary Report on the Worms (mostly parasitic) Collected in Lake St. Clair, in the Summer of 1893. Bull. Mich. Fish Com., 4: 49-54, 1 table.

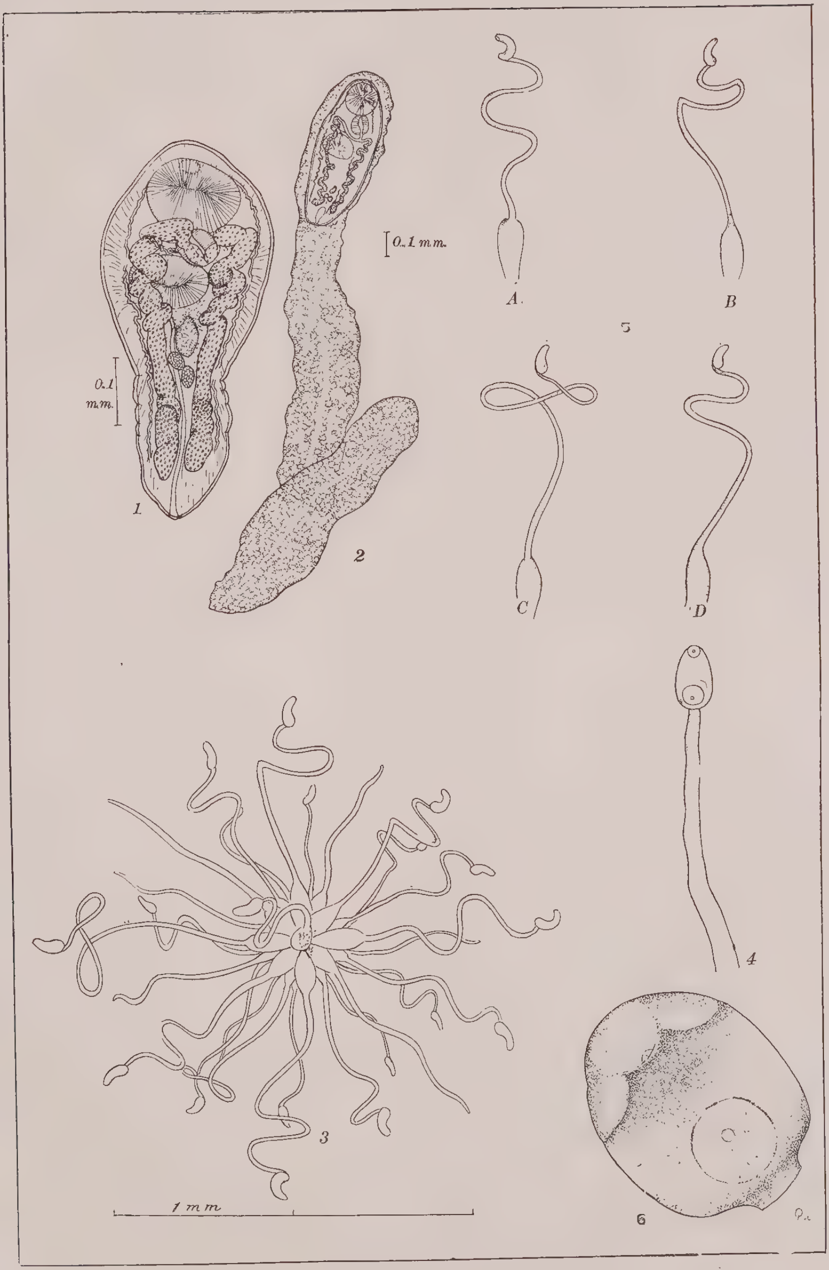
Wright, R. R. 1885. A Free-Swimming Sporocyst. Amer. Nat., 19: 310-311.

EXPLANATION OF PLATE

Figs. 1 and 2.—*Cercaria anchoroides.* 1, young distome just set free, $\times 37$; 2, *Cercaria* complete, $\times 105$.

Figs. 3 to 6.—*Cercaria gorgonocephala.* Free hand sketches from life. For details see text.

PLATE



ON THE ANATOMY AND RELATIONSHIPS OF SOME NORTH AMERICAN TREMATODES *

HORACE W. STUNKARD

As the result of an extended study of three families of North American trematodes, Polystomidae, Aspidogastridae and Paramphistomidae, certain points of interest in regard to the structure and classification have been elucidated. Since the publication of the completed work may be delayed, a brief statement of the more important points is presented here in advance of the appearance of the extended paper.

In the latest classification of the monogenetic trematodes, or Heterocotylea as they were termed by Monticelli, Odhner (1912) divided the group into two suborders, Monopisthocotylea in which a "true vagina is present," and Polyopisthocotylea in which a true vagina is wanting and the so-called "ductus vaginalis" is present. After careful study of the female ducts in the Polystomidae, I am able to show that the organ which functions as a vagina is homologous in all monogenetic trematodes and that there can be no division of the group on the basis of differences in this structure. In the complete paper the full evidence is submitted to show that the "true vagina" of the Monopisthocotylea is homologous to the originally single, secondarily paired and subsequently fused vaginae of the Polyopisthocotylea; altho the two suborders of Odhner are nevertheless valid, the essential difference between them is that the genito-intestinal canal is lacking in the former and present in the latter group.

The species that have been included in the genus Polystoma show a wider range of structural variation than is usually present in a natural genus. There are marked differences in the character of digestive and reproductive systems and variation exists also in the type of adhesive apparatus. In *P. integerrimum* the ceca are much branched, ramifying thru the body and caudal disc. In *P. alluaudi* the ceca occupy the same location but are merely lobed and have no secondary branches. In *P. bulliense,* according to Johnston (1912), "a diverticulum from the buccal cavity runs backwards, ventral to the pharynx, and for a distance equal to its length forming a median unpaired buccal pocket." In all other known species there is a simple bifurcate intestine, the ceca terminating just anterior to the caudal disc. In two specimens of *P. hassalli,* however, the ceca are connected posteriorly.

---

* Contributions from the Zoological Laboratory of the University of Illinois under the Direction of Henry B. Ward, No. 72.

The testis is a much branched structure in *P. kachugae,* in *P. integerrimum* it is lobed, and in the other known species it is oval or spherical. In *P. integerrimum* and *P. bulliense* the lateral vaginal swellings are formed by a large number of papillae which are perforated by fine canals, and in all other known species the vaginae are large open funnels and the lateral swellings are reduced or absent. In *P. integerrimum, P. bulliense* and *P. alluaudi* there is a long uterus which forms many loops in the intra-cecal area and contains a large number of eggs. In all other known forms, the uterus is situated at the level of the ovary on the opposite side of the body and contains a single large egg or embryo.

The caudal disc bears on its ventral face the chief organs of attachment. These consist of suckers and hooks, the former arranged in pairs, three suckers on either side of the median line. In all previously reported forms except *P. alluaudi,* the anterior suckers are separated by considerable distance giving the disc the shape described by Leidy as cordiform. In the single specimen of *P. alluaudi* described by Beauchamp, both the caudal and cephalic suckers are separated while those of each side are contiguous. In *P. orbiculare* n. sp. each sucker of the disc is separated from the two adjacent to it by uniform distances, making a perfect circle of bothria. In six species studied by the writer these suckers are complicated structures set more or less deeply in the parenchyma of the caudal disc. Their structure, character of insertion and muscular attachments are described in the complete paper. The caudal disc typically bears eighteen hooks. The larval hooks are anchor shaped and are situated six in a row between the anterior suckers, one inside each sucker at the base, and two or four between the posterior suckers. Between the posterior suckers there is also a pair of great hooks several times the size of the larval hooks, and in species in which a single pair of larval hooks is present, there is a third pair of hooks similar in shape to the great hooks and intermediate in size between the great and larval hooks.

In *P. orbiculare* n. sp. neither pair of the great hooks are present, and in *P. opacum* n. sp. there is a single pair of great hooks, very small and poorly developed.

The present study of the polystomes has emphasized the morphological variation and wide geographic distribution represented by the genus. This may mean either that the group is very old and has been subjected to conditions producing wide variation, or that it really lacks generic entity and consists of various heterocotylean forms which have specialized in the direction of an endoparasitic habit and that the morphological resemblance is cenogenetic.

Four new species are added to the genus Polystoma, as follows:

POLYSTOMA ORBICULARE *nov. spec.*

Length 2.7 to 3.75 mm.; width 0.9 to 1.2 mm. Caudal disc circular, 0.8 to 1.07 mm. in diameter, bothria arranged symmetrically in a circle. Only hooks present on disc are larval hooks in bases of suckers. Anterior sucker 0.25 to 0.27 mm. in length, 0.37 to 0.42 mm. in width; pharynx spherical 0.24 to 0.28 mm. in diameter; esophagus short. Testis spherical or oval, 0.36 to 0.5 mm. in length, 0.29 to 0.39 mm. in width, near or slightly anterior to middle of body. Genital coronet of 16 hooks equal in length. Ovary lateral, on either side of body, comma-shaped, 0.1 to 0.14 mm. wide by 0.14 to 0.185 mm. long. Vitellaria occupy dorsal and lateral areas from pharynx to caudal disc except in region dorsal to germ glands where they are reduced or absent.

In the urinary bladder of *Pseudemys scripta* from Raleigh, N. C., and of *Chrysemys marginata* from Chicago, Illinois, and Creston, Iowa.

POLYSTOMA OPACUM *nov. spec.*

Length 3.25 to 4 mm.; width 0.8 to 1 mm. Anterior sucker 0.2 to 0.22 mm. long, 0.23 mm. wide; pharynx spherical, 0.3 mm. in diameter; esophagus short. Testis spherical or oval, 0.4 to 0.5 mm. in diameter, slightly anterior to middle of body. Genital coronet of 32 similar hooks. Ovary lateral, comma-shaped or ovoid, 0.16 to 0.2 mm. long, 0.08 to 0.12 mm. wide. Vitellaria strongly developed, extend from pharynx to caudal disc, occupying lateral and dorsal regions of body except area over testis, ovary and uterus.

In esophagus of *Trionyx ferox* and *Malacoclemmys leseurii* from Newton, Texas.

POLYSTOMA MEGACOTYLE *nov. spec.*

Length 2.5 to 2.7 mm.; width 0.71 to 0.78 mm. Caudal disc cordiform; bothria large, overlap. Anterior sucker 0.28 mm. long, 0.35 to 0.42 mm. wide; pharynx 0.35 to 38 mm. long, 0.38 to 0.44 mm. wide. Testis near middle of body, 0.28 to 0.33 mm. long, 0.33 to 38 mm. wide. Genital coronet contains 36 hooks in one and 42 in another mounted specimen. Ovary broad comma-shaped organ on either side of median line, 0.1 mm. long, 0.075 mm. wide. Vitellaria extend in lateral and dorsal areas of body from pharynx to caudal disc, reduced or absent in small field dorsal to germ glands.

From oral cavity of *Chrysemys marginata,* Creston, Iowa.

POLYSTOMA MICROCOTYLE *nov. spec.*

Length 3 mm.; width 0.78 mm. Caudal disc cordiform; bothria small, separated. Anterior sucker 0.2 mm. long, 0.42 mm. wide; pharynx 0.37 mm. long, 0.4 mm. wide. Testis slightly anterior to

middle of body, 0.36 mm. long, 0.42 mm. wide. Genital coronet of 32 hooks, equal in length. Ovary lateral, 0.075 mm. long, 0.1 mm. wide. Vitellaria well developed, same extent as in *P. megacotyle*.

From oral cavity of *Chrysemys marginata*, Creston, Iowa.

In the family Aspidogastridae the three North American species have been restudied. A detailed comparison of specimens of *Aspidogaster conchicola* with the descriptions of Voeltzkow (1888), Stafford (1896), and other writers confirms former observations and substantiates the statements of Leidy (1851) and subsequent authors that *A. conchicola* occurs in this country. The examination of specimens of *Cotylaspis insignis* and *Cotylaspis cokeri* corrects and supplements former descriptions. Nickerson's (1902) classification of the family is revised and brought to date.

Representatives of three species of paramphistomes have furnished the basis for studies on that family. Two species are from North American turtles and the third is from a duck, *Anas platyrhynchos*. An examination of the literature showed that these forms could not be included in any previously described genera.

A new genus Alassostoma is created to contain the two species from turtles. The genus is characterized by the presence of large oral evaginations which open separately from the oral sucker, an esophageal bulb composed of concentric muscle lamellae, germ glands situated near the middle of the body in the median line, both testes anterior to the ovary, vitellaria consisting of small scattered follicles in the lateral and posteriorly in the median area of the body; Laurer's canal opens in the mid-dorsal line, anterior to the opening of the excretory vesicle; cirrus sac and uterus open to the exterior thru a common hermaphroditic duct. *Alassostoma magnum* n. sp. is taken as type of the genus in which is included also *Alassostoma parvum* n. sp.

The genus Alassostoma has the type of lymph and excretory systems present in the genus Schizamphistoma and designated by Looss (1912) as characters of the subfamily to which that genus belongs. Looss predicted that with the discovery of other genera it would be necessary to create a new subfamily to contain them, and at that time stated the subfamily characters. With the discovery of a second genus so similar to Schizamphistoma, the formal erection of the new subfamily is necessary. Schizamphistoma Looss was designated as the type genus and the name of the subfamily becomes Schizamphistominae. The distinguishing characters of the subfamily are stated by Looss to be two long excretory vesicles which extend singly to the anterior end of the body and a lymph system composed of three canals on either side of the body which extend longitudinally and

break up into many sinuses in the region of the suckers. The subfamily contains the genera Schizamphistoma, including also *S. spinulosum* which as indicated by Looss should probably be made the type of a new genus, and the genus Alassostoma.

Alassostoma magnum agrees with *Schizamphistoma scleroporum* in general appearance and size, in type of excretory and lymph systems, in character of vitellaria, and in general type of reproductive and alimentary organs; but *A. magnum* has large oral evaginations, which pockets are reduced and do not extend outside the sucker in *S. scleroporum;* further *A. magnum* lacks the preoral sphincter which is present in *S. scleroporum*. In *A. magnum* the uterus and cirrus sac open to the surface thru a common hermaphroditic duct; in *S. scleroporum* they open separately. In *A. magnum* the testes are further porteriad and the ovary is situated one fourth to one third of the body length from the posterior end instead of at the level of the anterior margin of the acetabulum as is the case in *S. scleroporum*. In the latter species the testes and ovary are widely separated whereas in *A. magnum* they are relatively close together. *A. magnum* agrees with *S. spinulosum* in the presence of oral evaginations and lack of preoral sphincter, but differs from it in the manner of coiling of the excretory vesicles, in the presence of common hermaphroditic duct, in the character of the vitellaria, as well as in relative positions of the testes and ovary. These morphological data show differences too fundamental to permit the inclusion of *A. magnum* in the same genus with either *S. scleroporum* or *S. spinulosum*.

Alassostoma parvum agrees with *A. magnum* in general morphological features, presence of oral evaginations, lack of preoral sphincter, type of lymph and excretory systems, in character of genital organs and ducts, also in relative position of testes and ovary. *A. parvum* therefore agrees with and differs from *S. scleroporum* and *S. spinulosum* in the same manner as *A. magnum*. That the two American forms are not different developmental stages of the same species is shown by the great difference in size of the worms and relative differences in size of suckers and genital organs. One mounted specimen of *A. magnum* 10 mm. long is not sexually mature while none of the individuals of *A. parvum* are more than 3 mm. in length. *A. magnum* is large with small suckers, whereas *A. parvum* is small with relatively large suckers; and this feature suggested the name Alassostoma.

Alassostoma magnum was collected from the large intestine of *Pseudemys troosti* and *P. elegans* from Havana, Illinois, and from *P. elegans* from Chicago, Ill. The specimens of *A. parvum* were found in the cloaca of *Chelydra serpentina* at Urbana, Ill.

The paramphistomes from *Anas platyrhynchos* were collected in Rock County, Nebraska. Unfortunately the fixation of the parasites is such that the excretory and lymph systems can not be traced, altho remnants of both appear in sections. This species closely resembles *Amphistoma lunatum* Diesing. Both are parasites of American ducks, and are the only paramphistomes at present known from avian hosts. They are nearly equal in size, are similar in shape, have a subterminal oral sucker, reproductive and digestive systems that compare closely, and acetabula of the same form, consisting of an anterior portion and a posterior overhanging lip which terminates on either side in a small cone-like projection. The species at hand differs from *A. lunatum* in its smaller oral evaginations, shorter esophagus, and in having oval, lobed testes and ovary instead of spherical germ glands. The acetabulum is nearer the ovary and the vitellaria are entirely extracecal while in *A. lunatum* they extend between the ceca.

Amphistoma lunatum has been placed as an appendix to every classification of the paramphistomes that has ever been attempted. With the discovery of a form so similar that the two must belong together, a new genus is proposed to contain the two species. The peculiar divided condition of the acetabulum suggested the name Zygocotyle for the genus. The species at hand, for which I propose the name *Zygocotyle ceratosa,* is designated as type and in the genus is included also the species *Z. lunatum* (Diesing). As diagnostic characters of the genus may be mentioned the subterminal oral sucker, posterior sucker divided or provided with a caudal overhanging lip, absence of cirrus sac, and separate openings for male and female ducts. Others will undoubtedly appear when the character of the excretory and lymph vessels are known. The genus Zygocotyle differs from all other known genera of the Paramphistomidae in the ventral position of the oral sucker and the peculiar character of the acetabulum. None of the existing subfamilies will include it fairly, but since the present classification of the Paramphistomidae is somewhat uncertain and the structure of the lymph and excretory systems of this genus is as yet unknown, no further attempt at classification of the group is made at this time.

Types of all the new species described in this paper have been deposited in the Helminthological Collection of the University of Illinois.

SUMMARY

Extended study of North American representatives of the three trematode families, Polystomidae, Aspidogastridae and Paramphistomidae has made possible the first comprehensive treatment in this

country of their structure and classification. Four new species are added to the genus Polystoma and three new species of two new genera are added to the Paramphistomidae.

Literature Cited

Johnston, S. J. 1912. On Some Trematode Parasites of Australian Frogs. Proc. Linn. Soc. N. S. Wales, 37: 285-362, pl. 14-43.

Leidy, J. 1851. Helminthological Contributions II. Proc. Acad. Nat. Sci. Phila., 5: 224-227.

Looss, A. 1912. Ueber den Bau einiger auscheinend seltener Trematoden-Arten. Zool. Jahrb., Suppl., 15: 323-366, 3 pl.

Nickerson, W. S. 1902. *Cotylogaster occidentalis* n. sp. and a Revision of the Family Aspidobothridae. Zool. Jahrb., Syst., 15: 597-624, 2 pl.

Odhner, T. 1912. Bemerkungen zum natürlichen System der Monogenen Trematoden. Zool. Anz., 39: 337-351.

1913. Noch einmal die Homologien der weiblichen Genitalwege der monogenen Trematoden. Zool. Anz., 41: 558-559.

Stafford, J. 1896. Anatomical Structure of *Aspidogaster conchicola*. Zool. Jahrb., Anat., 9: 477-542, 4 pl.

Voeltzkow, A. 1888. *Aspidogaster conchicola*. Arb. zool.-zoot. Inst. Würzb., 8: 249-292, 5 pl.

DAUERCYSTFORMATION OF *TRICHOMONAS INTESTINALIS* *

Kenneth M. Lynch, Charleston, S. C.

Ucke (1908), Bohne and Prowazek (1908), and Bensen (1910), have described encystment of *Trichomonas intestinalis.* Bensen (1910) has also described an encystment for *Trichomonas vaginalis* differing from that of *Trichomonas intestinalis,* and Dobell (1908) reports a dauercyst of *Trichomonas batrachorum.* Alexeieff (1911) disputes the nature of the so-described cyst of *Trichomonas intestinalis,* asserting that it is in reality an ascomyces, a vegetable organism akin to the yeasts, and proposes for it the name *Blastocystis enterocola.* Wenyon (1905) and others uphold Alexeieff's contention, Wenyon calling the organism *Blastocystis hominis.*

The question of encystment of *Trichomonas intestinalis* is an interesting one to me because of the questioned nature of the cyst which has been previously described and because of the prevalence of the parasite in this community. For several years I have been observing Trichomonas as a parasite of several locations in the human body and in certain lower animals, and have frequently encountered a form which has proven to be a distinct cyst and not to be confused with that previously described as an encysted *Tr. intestinalis* by Ucke, Bohne and Prowazek, and Bensen, and as *Blastocystis enterocola* by Alexeieff.

At the present I have under observation a man who furnishes this cyst in large numbers and with distinct characteristics. This man is a negro who is in the hospital with chronic endocarditis and who gives no history of dysentery.

In the stool no other protozoon has been found, no cell corresponding to the previously described *Tr. intestinalis* cyst and none of the Blastocystis of Alexeieff. There are many active Trichomonas conforming to the typical organism, averaging about 8 by 12 micra in size, of elongated pear shape, with constant undulating membrane, three flagella, the stiff spine projecting posteriorly, vacuolated cytoplasm with numerous ingested bacteria, and with nucleus indistinct but showing well in stained preparations. On exposure the active form soon becomes inactive, stationary, and does not develop the irregular ameboid, non-flagellated, undulating form. It is one of the most fixed types I have observed.

---

* From the Department of Pathology and Research Medicine of the Medical College of the State of South Carolina.

The encysted form is almost as numerous as the active and commonly exhibits a tendency to occur in pairs. It is about three-fourths the size of the active, of a typical pear shape, and has a transparent shell of uniform thickness. The enclosed parasite has a regular ovoid contour and a finely granular grayish appearance. On one side nearer to the small end the nucleus is visible as a refractive granule, and on the other the undulating membrane is seen as a refractive wavy line extending from end to end.

More definite observations of this cyst and its different structures may be made from stained specimens. The technic of staining which I have used is as follows: A thin spread of the feces while still wet is fixed in warm saline alcoholic corrosive sublimate for ten minutes, placed in absolute alcohol ten minutes, iodin solution ten minutes, alcohol ten minutes, washed in distilled water, mordanted in 4% aqueous ferric ammonia alum overnight, stained in 1% alcoholic hematoxylin one day, decolorized in 2% aqueous ferric ammonia alum, counterstained in alcoholic eosin and cleared in carbol-xylol.

In such a preparation the encysted form is more deeply stained than the active. It is about 6 by 8 micra in size and of perfectly symmetrical pear shape. The anterior end projects on a shoulder slightly beyond the general line; the wall is uniformly distinct; and the space between the cyst wall and the body of the parasite is usually distinct and clear, it being usually broader at the anterior end.

The body of the enclosed parasite is of symmetrical, ovoid shape, slightly pointed anteriorly, of dull brick-red color, finely granular and contains no vacuoles or food particles. A fine dark line beginning as a granule in the anterior end runs directly backward to the posterior end. This I take to be the stiffening rib of the undulating membrane because of its close association in origin and termination with that organ. The undulating membrane is distinct as a darker line beginning in close connection with this rib and extending backward along one side of the body in a wavy course to the posterior end, where it curves around the extremity of the parasite and comes to end near this end of the rib. The nucleus is comparatively large, of ovoid form, but lies farther back than in the active parasite and on the side opposite to the undulating membrane. Its usual position is in the posterior part of the anterior half, and between the midline and the body wall. It has a distinct dark-stained rim and a large chromatin mass. This chromatin usually occurs as an irregular black body almost filling the nucleus, but is in some broken into smaller granules and in others distributed around the inner edge of the nuclear rim. In addition to the undulating membrane the cyst usually shows two or three more delicate lines arising in close association with that organ and passing backward over the body for about two thirds of its length. These are

probably flagella. They stain poorly and are not constantly seen, especially in the more faintly stained specimens. The characteristics of the fully formed cyst may be seen in Figure A3 which is an offhand drawing.

In addition to this fully developed cyst which predominates, there are young cysts and forms of apparent pre-encystment. There is a form which is smaller than the active parasite, shorter and more blunt, with cytoplasm somewhat reticulated, but showing no vacuoles and no bacteria or other ingested materials, and with nucleus more distinct and with larger amounts of chromatin. This I take to be a pre-encysted stage. Figure A1 appears to be a young cyst before the wall has reached full formation. Its shape is typical; the body wall is thick; and the internal organs appear as in the fully encysted. A further stage appears to be the form in which there is a space between the shell and parasite only at the anterior end, these parts being in immediate contact around the remainder of the body, see A2.

Fig. A1, 2, and 3.—Drawings of different stages of dauercysts of *Trichomonas intestinalis* as seen in specimens stained in hematoxylin.

Further development suggestive of multiplication I have not seen in these cysts; and the preservation of the undulating membrane and flagella, together with the single nucleus, indicate that the process is not for reproduction but simply for resistance.

DISCUSSION

According to these observations the formation of a resistance cyst plays a part in the life of *Trichomonas intestinalis* as occurs with other intestinal protozoa, and I believe is the stage in which the parasite may be transmitted. That infection by the contracted but non-encysted parasite occurs I am not prepared to dispute, and it seems probable to me that the contracted form may be a pre-encystment stage. Reasoning by analogy leads one to believe that infection through the stomach by means of the non-encysted is not probable, and certain observations which I have made of the purely active Trichomonas from the vagina also rule against such a manner of infection.

I have previously reported (1915) a case of vaginal and mouth infection by Trichomonas which did not infect the intestine, this being determined by repeated examination after purging. I have since had a similar case of trichomoniasis of the vagina and mouth; and again by repeated antemortem and also postmortem examinations failed to find the intestine infected. In these cases the parasites seemed identical in the two situations, and there was never any but the pure fixed type of active form. There were enormous numbers in the mouth for

Fig. B1, Dauercyst of *Tr. batrachorum* by Dobell; 2, Encysted *Tr. vaginalis* by Bensen; 3, Cyst of *Tr. intestinalis* by Wenyon; 4, Encysted *Tr. intestinalis* by Ucke; 5, Encysted *Tr. intestinalis* by Bohne and Prowazek; 6, Encysted *Tr. intestinalis* by Bensen; 7 and 8, *Blastocystis enterocola* by Alexeieff; 9, *Blastocystis hominis* by Wenyon.

considerable periods of time; and if the active Trichomonas is capable of transmission through the stomach to the intestine in man, the swallowing of these organisms should have produced an intestinal infection in these women.

It is not my purpose at this time to enter into a discussion as to the nature of the previously described encysted *Tr. intestinalis* which has been called Blastocystis by Alexeieff; but from extensive observa-

tion of both organisms, both associated and occurring separately, I am in accord with the view that it is not a Trichomonas cyst.

In order that the lack of resemblance of this cell to the cyst here described may be seen I have included copies of figures by Ucke, Bohne and Prowazek, Bensen, Alexeieff, and Wenyon. Figures B5 and 6 are from a hematoxylin-stained specimen. Hence the main difference in appearance to Figures B4, 7, 8, and 9, which are representations of the unstained cell, since the internal part of the cell, which is often transparent in the fresh specimen, stains rather deeply and the nuclei are more distinct in the stained. Figures B4, 5, and 6 are illustrations of the so-called *Trichomonas intestinalis* cysts of Ucke, Bohne and Prowazek, and Bensen. Figures B7, 8, and 9 are illustrations of Blastocystis by Alexeieff and Wenyon. There is seen to be no point of similarity between the Trichomonas cyst of Ucke, Bohne and Prowazek or Bensen and that here described and pictured in Figure A; whereas, barring differences in preparation and in stage of the organism, the Blastocystis of Alexeieff and Wenyon and the Trichomonas cyst of Ucke, Bohne and Prowazek and of Bensen are apparently one and the same organism.

There is however some resemblance between this cyst and Bensen's encysted *Trichomonas vaginalis* (Figure B2) and Dobell's cyst of *Trichomonas batrachorum* (Figure B1), which I have taken the liberty of copying for comparison. In the last two the flagella are preserved until the cyst is formed, but lost afterwards; while the typical shape of the *Trichomonas intestinalis* cyst is not observed, and the undulating membrane and axostyle are not seen. The nucleus is also different, in that in *Tr. intestinalis* it is more rounded and placed more posteriorly, while in both *Tr. batrachorum* and *Tr. vaginalis* it is forward, larger and spindle shaped. Then in the case of *Tr. vaginalis* Bensen has described and illustrated further development of the cyst for multiplication, while further development of the cyst here described has not been seen.

CONCLUSION

Accordingly, therefore, the formation of a resistance cyst does play a part in the life of *Trichomonas intestinalis,* and this cyst bears no relationship to the cell which has been previously described as an encysted *Tr. intestinalis* by Ucke and others, and as *Blastocystis enterocola* by Alexeieff, and differs essentially from Dobell's dauer-cyst of *Trichomonas batrachorum* and Bensen's encysted *Tr. vaginalis.*

SUMMARY

Encystment of *Trichomonas intestinalis* has been investigated for many years. That which has been previously described has not stood

the test of investigation. The morphology of the cyst here described identifies it with the parasite from which it arises. It is not to be confused with any cell occurring in the intestine and feces. Whether this cyst releases more than one organism or whether *Trichomonas intestinalis* has a multiplication cyst remains an unanswered question.

References Cited

Alexeieff, A. 1911. Sur la nature des Formations Dites "Kystes de *Trichomonas intestinalis.*" C. R. Soc. Biol., 71: 296-298.

Bensen, W. 1910. Trichomonas intestinalis und vaginalis des Menschen. Arch. Protist., 18: 115-127.

Bohne, A., und Prowazek, S. V. 1908. Zur Frage der Flagellatendysenterie. Arch. Protist., 12: 1-8.

Dobell, C. C. 1908. *Trichomonas batrachorum* Perty. Proc. Cambridge Philos. Soc., 14: 428-433.

Lynch, K. M. 1915. Trichomoniasis of Vagina and Mouth. Amer. Jour. Trop. Diseases and Prev. Med., 2: 627-634.

Ucke, A. 1908. Trichomonaden und Megastomum im Menschendarm. Centralb. Bakt. Par., I. Abt., 45: 231-233.

Wenyon, C. M. 1905. The Common Intestinal Protozoa of Man: Their Diagnosis and Pathogenicity. The Lancet, London, 189: 1173-1183.

NOTES ON TWO CESTODES FROM THE SPOTTED STING-RAY

Edwin Linton

A single specimen of a species of cestode found in the spiral valve of a cow-nosed ray (*Rhinoptera bonasus*) at Woods Hole, July 29, 1887, was made the type of a new genus and species (*Tylocephalum pingue*). No other examples of this genus have been found at Woods Hole, but on June 30, 1908, at the Tortugas laboratory, I obtained two specimens of a cestode from the spotted sting-ray (*Aetobatis narinari*) which are to be referred to the genus *Tylocephalum*. The specimen from the cow-nosed ray was a less mature strobile than those from the spotted ray; a comparison of the genitalia, therefore, cannot be made. There appears to be enough difference, however, in other particulars to justify referring the Tortugas specimens to a new species. While both hosts belong to the family of eagle rays, there is enough difference between them in the way of geographical range and generic features to make it unlikely that the same species of cestodes should be found in each.

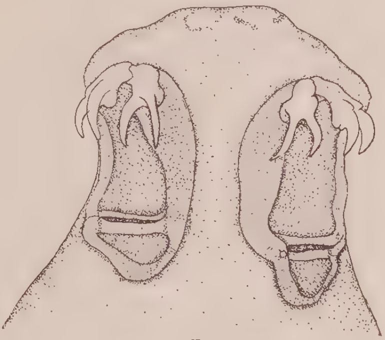
TYLOCEPHALUM MARSUPIUM *nov. spec.*

Scolex: The relatively large, muscular portion (myzorhynchus) is subglobular, its length in a living specimen 0.16 and breadth 0.21 mm.; bothria united into a subglobular disc with four auxiliary acetabula, length of disc 0.30, breadth 0.69 mm. The constriction noted in the Woods Hole specimen not present. As in the case of the specimen from the cow-nosed ray, the scoleces were rather firmly fastened to the mucous membrane of the spiral valve. One of the worms was fixed without detaching it, and was sectioned together with a small piece of the intestinal wall. The sections show that the myzorhynchus alone had entered the mucous membrane.

Strobile: The segments begin nearer the scolex than they do in *T. pingue*. Just behind the scolex, where the breadth was 0.16, the strobile was crossed by crowded lines. One-half millimeter back of the scolex the well-defined segments were 0.014 mm. in length and 0.18 mm. in breadth. Three millimeters back the length of the segments is about 0.05 and the breadth 0.24; ten millimeters back the length is 0.12, the breadth 0.28; twenty millimeters back the length is 0.28, the breadth 0.24; thirty millimeters back the length is 0.46, the breadth 0.28; forty millimeters back of the scolex the length is 0.56, the breadth 0.38 mm. The last segments are somewhat variable in their dimensions, but are about one millimeter in length and 0.5 mm.

in greatest breadth. They are vase-shaped, constricted at the anterior end, swelling out to the maximum breadth behind the middle, slightly constricted near the posterior end with a moderately projecting posterior margin. One proglottis had the following dimensions: Length 0.84; breadth, anterior 0.21, maximum 0.56, posterior 0.39 mm. The strobile is especially distinguished by the strongly developed longitudinal muscles. The longitudinal muscles are disposed in radial bundles near the scolex (Fig. 1), but farther back lie in a well-defined zone (Fig. 2). In segments in which the genitalia have become differentiated this zone of muscle bundles coincides in position with the vitellaria (Fig. 4).

A



B

Text Fig. A.—*Tylocephalum marsupium.* View of scolex in life, somewhat flattened and seen from behind. Breadth of scolex 0.7 mm.

Text Fig. B.—*Onchobothrium tortum.* Side view of scolex; balsam. Diameter at base of hooks 0.64 mm.

Genitalia: The general plan of arrangement of the genitalia is shown in Figure 7. The vitellaria are peripheral and consist of rather finely granular masses lying between and also centrally to the muscle bundles. The testes are in the median region. In the younger proglottids they occupy most of the interior, but as the proglottids mature they give way to the seminal receptacle and ovary. The cirrus-pouch is relatively small and oval, opening near the margin not far from the middle of the length. The vagina opens into the genital cloaca, passes along one side of the cirrus pouch, becomes more or less convoluted and expands into a capacious seminal receptacle. This was filled with spermatozoa in all the later proglottids. The ovary is lobed and is situated at the posterior end of the proglottis.

The uterus was still rudimentary even in the mature proglottids. In a section a small cluster of minute bodies was seen. They lay in the lumen of the uterus, were yellowish brown, and about 0.010 by 0.007 mm. in the two principal diameters.

ONCHOBOTHRIUM TORTUM *nov. spec.*

Ten specimens of this form were obtained from a spotted sting-ray (*Aetobatis narinari*), June 30, 1908. The scolices were imbedded in the intestinal wall and had caused some ulceration. One of the worms, straightened out on a glass plate in sea water, measured 220 mm. in length. Anterior end sub-cylindrical, with a tendency to coil spirally; color dark ashy-gray. Scolex long-clavate, armed with four pairs of short, sharp, two-pronged hooks. Each pair of hooks situated at the anterior end of one of the four bothria. The latter are oblong, trough-shaped, with two costæ near the posterior end. Behind the scolex the body is at first sub-cylindrical and crossed by fine, closely crowded lines for a considerable distance. The segments outlined by these transverse lines remain closely crowded, while the adult proglottids begin rather abruptly. The average length of the first 12 adult proglottids was 0.8 mm., the breadth being about the same or slightly greater. The diameter of the sub-cylindrical portion of the strobile was about 1.5 mm. The scolex and anterior portion of the strobile are much thicker than the adult proglottids. Diameter of scolex, in alcohol, anterior 0.85, middle 0.77; diameter of neck, a short distance back of the scolex, 1.4 mm. Dimensions of one of the posterior proglottids: life, length 1.47; breadth, anterior 0.5; middle 0.8, posterior 0.6 mm. Dimensions of scolex mounted in balsam: length 0.97; breadth, at base of hooks, 0.97, behind hooks, 0.81; breadth of neck, a short distance behind the scolex, 1.27 mm. In the mounted specimen the neck is seen to be traversed by strong longitudinal muscle bundles which are closely crowded together, each bundle about 0.06 mm. in diameter. About 16 bundles were counted near the head; farther back they are divided into a larger number of smaller bundles. Two spiral vessels show distinctly in the mounted specimen. The strobile narrows as the proglottids become distinct. In the specimen which measured 220 mm. there were distinct and well-formed segments on the last 150 mm. The maturing segments were at first much broader than long, then squarish, then longer than broad, the last ones three times as long as broad. The posterior margins of the proglottids project slightly and have crenulate borders. One of the posterior proglottids of a mounted strobile has the following dimensions: length 1.86; breadth, anterior 0.36, constriction near anterior end 0.25, middle 0.40, posterior margin 0.54 mm. The genital apertures are marginal at about the middle of the length. They are irregularly alternate. No ova were seen.

The general plan of arrangement of the genitalia is shown in Figure 8. The cirrus is armed with slender, bristle-like spines; a few folds of the vas deferens are included in the oval cirrus-pouch at its

medial end. The voluminuous folds of the vas deferens form the seminal vesicle and occupy the median third of the anterior half of the proglottis. The testes are situated in the anterior half of the proglottis, and occupy the median region on each side of the vas deferens. On the marginal sides of the testes are the vitelline glands which extend along each marginal border of the entire length of the proglottis, being interrupted only at the point where the cirrus pouch and the accompanying vagina approach the genital aperture. The uterus was represented by a tubular structure lying along the median line near one of the lateral faces of the proglottis, and extending from nearly one end of the proglottis to the other. The ovary is a lobed organ and fills all the space between the marginal vitellaria behind the cirrus pouch. The vagina opens at the genital pore immediately in front of the cirrus and lies alongside the anterior border of the cirrus pouch. At this point it is thick-walled and glandular. It becomes tubular at about the level of the median end of the pouch and passes along the median line beneath the uterus to about the middle of the ovary. The relative positions of vagina and uterus are shown in Figure 9, which is sketched from a transverse section of a maturing segment at a level which passes very near the genital aperture, shows a portion of the vagina near the margin, cuts into some folds of the vas deferens, and passes thru the vagina again near the middle of the segment, where it lies on the medial side of the uterus. The section also catches a few of the anterior lobes of the ovary. In this section the characteristic longitudinal muscles are seen as an inner circle of larger and an outer circle of smaller bundles. The lateral vitellaria and the median testes flanking the folds of the seminal vesicle are also shown .

SUMMARY

Two new species of cestodes, of the genera *Tylocephalum* and *Onchobothrium,* respectively, are described in this paper. One of them, *T. marsupium,* is the first cestode of this genus to be recorded since the genus was established in 1887. Thus far representatives of this genus have been found only in the eagle rays.

Altho the two genera belong to quite different families, they possess an interesting feature in common in the strongly fasciculated longitudinal muscle layers. Both species were fastened to the mucous membrane of the spiral valve which, at the point of attachment of the onchobothria, was somewhat ulcerated.

EXPLANATION OF PLATE

Fig. 1.—*Tylocephalum marsupium.* Transverse section of neck. Diameter 0.22 mm.

Fig. 2.—*Tylocephalum marsupium.* Transverse section of early proglottis, showing rudiment of genitalia and peripherally arranged longitudinal muscle bundles. Greater diameter 0.65 mm.

Fig. 3.—*Onchobothrium tortum.* Transverse section of neck, showing longitudinal muscle bundles and vessels of the vascular system. Longer diameter of section 1.12 mm.

Fig. 4.—*Tylocephalum marsupium.* Transverse section of mature proglottis in front of cirrus bulb; longer diameter 0.45 mm.

Fig. 5.—*Onchobothrium tortum.* Longitudinal view of neck showing muscle bundles. Breadth 1.17 mm.

Fig. 6.—*Onchobothrium tortum.* View of retracted cirrus, and vagina; from longitudinal section.

Fig. 7.—*Tylocephalum marsupium.* Posterior proglottis; outline from life; genitalia partly diagrammatic. Length 0.8 mm.

Fig. 8.—*Onchobothrium tortum.* Posterior proglottis; balsam. Length 1.6 mm.

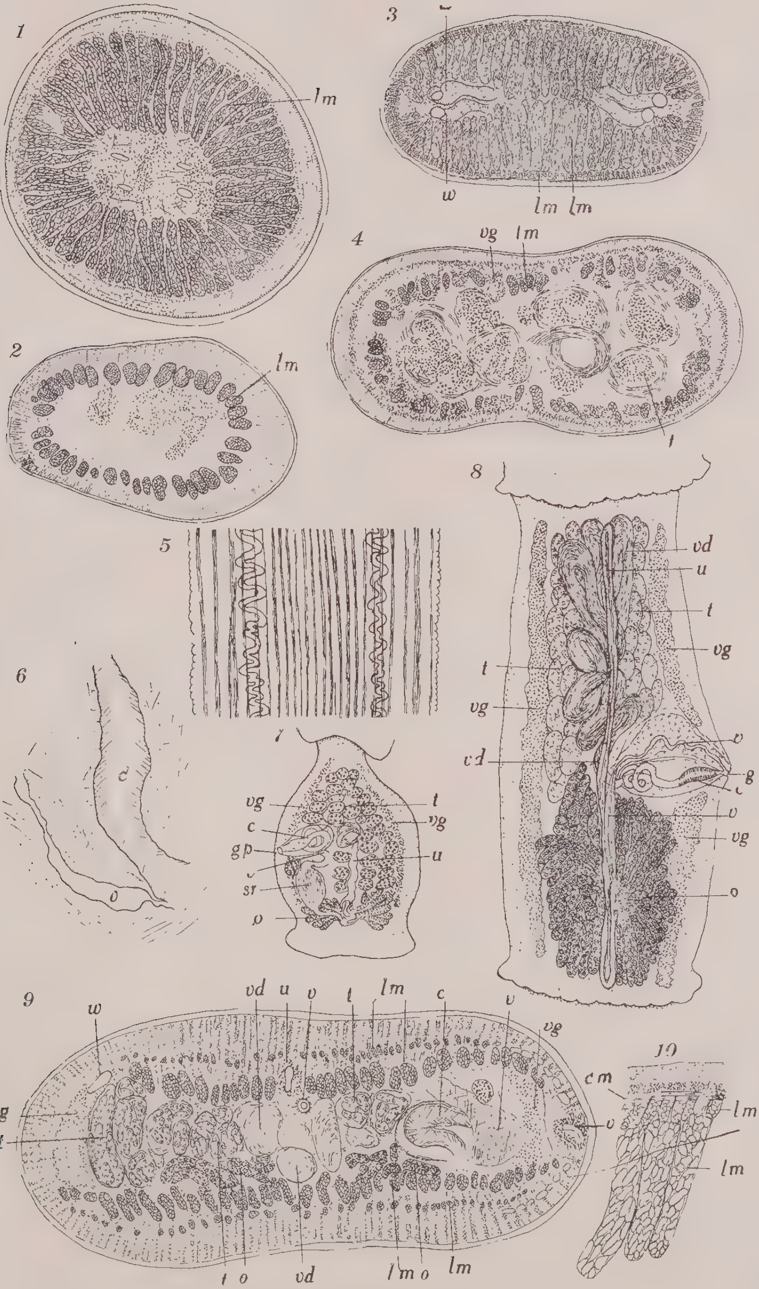
Fig. 9.—*Onchobothrium tortum.* Transverse section of a somewhat younger proglottis than that shown in Figure 8. Longer diameter of section 1.12 mm.

Fig. 10.—*Onchobothrium tortum.* Details of musculature.

Abbreviations Used

c, retracted cirrus
cm, circular muscle layer
gp, genital pore
lm, longitudinal muscle bundles
o, ovary
sr, seminal receptacle
t, testes
u, uterus
v, vagina
vd, vas deferens
vg, vitellaria
w, longitudinal vessel

PLATE



1
lm
3
w
lm lm
2
lm
4
vg lm
t
5
6
c
o
7
vg
t
vg
c
gp
u
sr
o
8
vd
u
t
vg
t
vg
v
cd
g
c
v
vg
o
9
w
vd u v
t
lm
c
v
vg
vg
t
v
t o
vd
lm o
lm
10
cm
lm
lm

A CASE OF THE OCCURRENCE OF *ASCARIS TRIQUETRA* SCHRANK IN DOGS *

A. C. Walton

While working on the spermatogenesis of certain Ascaridae last year, I found that the chromosomes of the ascarids from dogs did not agree with those of the ascarids from the dog as given by Kultschitzky (1888) and by Marcus (1906) either in number, behavior, or the presence of an idiochromosome group. The work of Glaue (1908, 1909, 1910) has shown conclusively that the ascarids of the dog and of the cat are anatomically distinct species, which should be designated respectively as *Ascaris canis* Werner, and *Ascaris felis* Goeze, and not merely varieties of *Ascaris mystax* Zeder. The work of Edwards (1911) on *A. felis* and that of Marcus (1906) on *A. canis* have given us conclusive evidence that these two forms are entirely dissimilar as to the number and the behavior of the chromosomes. From these taxonomic and cytological proofs, the long mooted question of the identity of the two varieties seemed definitely settled; but the apparent contradiction in the gametogenesis of the dog ascarids shown by my discovery seemed to me sufficient to warrant the reopening of the question. If the number and behavior of the chromosomes in *Ascaris canis* were similar to the number and behavior of those in *Ascaris felis,* the two forms might be in fact only sub-species; varying taxonomically owing to their different environments.

The results of my study are contained in a paper now in press, the taxonomic work of which showed that the species with which I was working were the ones recognized by helminthologists as the usual inhabitants of the intestine of the dog and the cat, respectively. My work on the gametogenesis of *A. felis* agreed with that of Edwards (1911) in showing nine chromosomes as the haploid number, one of which is a member of an X-Y idiochromosome pair.

Marcus (1906) has shown that what he called *A. canis* has ten paired and two unpaired tetrad chromosomes as the diploid number. From his description it seems probable that these two unpaired chromosomes act as members of an X-Y idiochromosome group, but he did not so call them. My work on the commonest parasite of the dog has shown that in the male there are thirty tetrad chromosomes as the diploid number, of which twenty-four are united in pairs, and the

---

* Contributions from the Zoological Laboratory of the Museum of Comparative Zoology at Harvard College, No. 283.

other six form a heterochromosome group of the X type. The female has thirty-six tetrads (eighteen di-tetrads) as the diploid number.

Private correspondence between Dr. S. I. Kornhauser, of Northwestern University, and Dr. Marcus has shown that the majority of the material upon which the latter based his work was *not* obtained from dogs, but came mostly from other members of the dog family and also from bears.

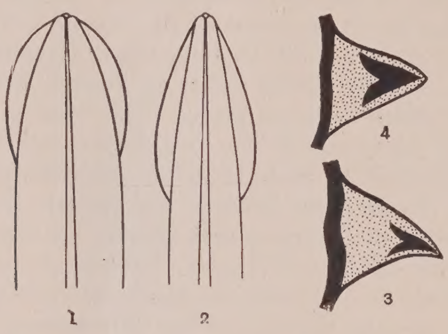


Fig. 1.—Dorsal aspect of the anterior end of *Ascaris triquetra* Schrank (× 25).

Fig. 2.—Same view of *Ascaris canis* Werner (× 25).

Fig. 3.—Cross-section. Posterior aspect of the right wing of *A. triquetra* (× 160).

Fig. 4.—Same for *A. canis* (× 160).

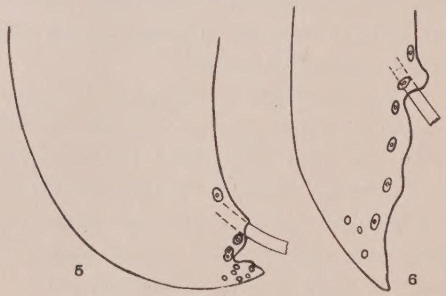


Fig. 5.—Lateral view of the right side of posterior end of male *A. triquetra* (× 25).

Fig. 6.—Same view of male *A. canis* (× 25).

All drawings were made with the aid of a camera lucida.

During the past two years I have been able to examine worms taken from twenty-five dogs, and of the total of two hundred worms thus obtained, all but two have answered taxonomically and cytologically to the type described above as the commonest Ascaris in dogs, i. e., *Ascaris canis* Werner. These two exceptional specimens, a male

and a female of the same species, differed considerably in taxonomy from the ordinary type of *Ascaris canis* Werner. The following table compares the main features of the two forms:

	A. canis Werner	*A. triquetra* Schrank
Length of male	120 mm	60 mm.
Length of female	220 mm	100 mm.
Shape of oral wing	Lanceolate	Broadly lanceolate
Thickness of oral wing	0.17 mm	0.18 mm.
Breadth of oral wing	0.165 mm	0.18 mm.
Length of oral wing	2.7 mm	1.9 mm.
Chitin rod of wing	Long and broad	Shorter and narrower
Post-anal papillae	7	8
Ventral row	4	4 (one double).
Dorsal row	3	4 (2 rows, 2 each)
Shape of tail of male	Slopes gradually to a point	Bends sharply ventrad to a short, blunt end

The comparison of the two species shows that the less common one agrees with *Ascaris triquetra* Schrank, which earlier writers believed to be synonymous with *A. mystax* Zeder and *A. marginata* Rudolphi. Marcus (1906) had identified his *A. canis* with the *A. marginata* studied by Kultschitzky (1888). Cytological examination of the sex cells of this *Ascaris triquetra* Schrank shows that there are twenty tetrad chromosomes, arranged in ten pairs, and also two unpaired tetrads, as the diploid number. This agrees with the facts recorded by Marcus for his material, and I believe, therefore, that the Ascaris studied by him was also *Ascaris triquetra* Schrank, known to Kultschitzky as *Ascaris marginata* Rudolphi.

My work, then, has shown that, while *Ascaris canis* Werner is the common parasitic nematode of the dog, *Ascaris triquetra* Schrank may be an inhabitant of the same dog that harbors individuals of the species *A. canis* Werner, though this occurs only rarely. It has also shown that the nematode studied by Marcus (1906) was probably *Ascaris triquetra* Schrank, rather than *Ascaris canis* Werner.

I wish here to express my obligation to Dr. S. I. Kornhauser for his notes and especially to Dr. E. L. Mark for his supervision of the preparation of this paper.

Bibliography

Edwards, C. L. 1911. The Sex-chromosomes in *Ascaris felis.* Arch. f. Zellforsch., 7: 309-313, Taf. 23-26.

Glaue, H. 1908. Zur Unterscheidung von *Ascaris canis* und *A. felis* (*Ascaris canis* s. *mystax*). Zool. Anz., 33: 785-790, 3 figs.

1909. Beiträge zu einer Monographie der Nematodenspecies, *Ascaris felis* und *Ascaris canis.* Zeit. f. wiss. Zool., 95: 551-593, 26 figs.

1910. Beiträge zur Systematik der Nematoden. Zool. Anz., 35: 744-759, 5 figs.

Kultschitzky, N. 1888. Ueber die Eireifung und die Befruchtungsvorgänge bei *Ascaris marginata.* Arch. f. mikr. Anat., 32: 671-682, Taf. 26-27.

Marcus, H. 1906. Ei- und Samenreife bei *Ascaris canis.* Arch. f. mikr. Anat., 68: 441-491, Taf. 29-30, 10 text figs.

REVIEWS AND NOTES

The staff of the Research Department at the Severance Union Medical College in Seoul, Korea, of which Dr. Ralph G. Mills is director, has undertaken a review for English readers of current periodicals in Japanese medical literature. This is printed every two months in the China Medical Journal, and also circulated separately. The publication is likely to be of great importance to parasitologists because of the activity in Japan at present in the investigation of diseases caused by animal parasites which play a great rôle in that country.

The first (?) number, dated 1916 and recently received, contains a review with illustrations of long articles on the development of the supposed last stage in the life history of Paragonimus by Nakagawa, on the first intermediate host of that parasite by the same author, and on an investigation of the Lungfluke in Korea by Kakami, in addition to numerous other items mostly pathological. The reviews are very well written and present valuable material not otherwise accessible to the American investigator.

It is with great sorrow that the JOURNAL announces the death of one of its collaborators, the distinguished German helminthologist, Max Lühe, Professor at the University of Königsberg, who died of wounds received in the war. The death of Lühe is a great loss to science and the world. His contributions to the literature of parasitology embrace important and extensive studies on Protozoa, Trematoda, Cestoda, Nematoda, and Acanthocephala. It is hoped to print at an early date a biographical sketch of Professor Lühe accompanied by a portrait.

Harvard University has issued the first formal announcement of the School of Tropical Medicine. Courses are open to graduates of recognized medical schools so that the work becomes a part of the Graduate School of Medicine. Dr. Richard P. Strong is Director of the School of Tropical Medicine and in the work announced are courses in protozoology, helminthology, and tropical entomology.